REMARKS/ARGUMENTS

Claims 1-6, 57-59 and 63-68 are pending in this application. Claims 15-21 were previously cancelled and claims 7-14 and 22-28 are canceled herewith without prejudice. Claims 29-56 and 60-62 were previously withdrawn due to a restriction requirement. Claims 1-6, 57-59 and 63-65 are currently amended for the purposes of clarity. Applicant has added new claims 66-68. No new matter has been added by any of these amendments.

Declarations under 37 CFR 1.132

Applicant respectfully requests the entry of the Declarations under 37 CFR 1.132, which were submitted with Applicant's response of May 3, 2004, as they are now timely submitted with respect to the RCE. Applicant further requests the entry of yet another declaration under 37 CFR 1.132 submitted herewith, in response to the Examiner's request for a description of the materials and methods of the declarant's experiments.

The Examiner states that it is unclear if the testing was *in vitro* or *in vivo*, however, it is noted that the previously submitted declaration of Dr. Preston explicitly stated that the experiments were obtained through *in vitro* testing. The presently submitted declaration further explains the experimental evidence in support of the nonobviousness of the instant invention over the prior art.

Furthermore, the Examiner states that the "declarations provide no clear side-by-side comparison with the closest prior art, i.e., comparing with Jukema et al." However, one of the previously submitted declarations was written by Dr. Jukema and explicitly states, "I did not present data nor discuss the notion that the antioxidant effect of the atorvastatin metabolite would be enhanced by amlodipine in a synergistic fashion...[or] a synergistic effect observed with amlodipine and atorvastatin metabolite could be reproduced using other stains." The Applicant believes this is a clear and direct comparison with the closest prior art (as discussed below).

Rejection of claims 1-14, 22-28, 57-59 and 63-65 under 35 USC § 103(a)

Claims 1-14, 22-28, 57-59 and 63-65 are rejected under 35 USC 103(a) as being unpatentable over Davison *et al.* (4,879,303) and Borge *et al.* (5,385,929) in view of Jukema (Arteriosclerosis, Thrombosis, and Vascular Biology, v16, No. 3 (1996), pp425-430) and Merck Index. Applicant respectfully disagrees.

The Examiner states that Davison teaches "a pharmaceutical composition comprising amlodipine besylate useful in treating ischemic heart disease ..., see claim 1 and abstract in particular." The Examiner continues, "Bjorge ... teaches a pharmaceutical composition comprising ... metabolites of atorvastatin and a pharmaceutical carrier useful in inhibiting cholesterol synthesis ... see claims 12, 18 and abstract."

The Examiner adroitly points out that "Davison ... and Bjorge ... taken together do not expressly disclose a [pharmaceutical] composition comprising amlodipine besylate and a hydroxylated metabolite of atorvastatin." Applicant completely concurs with this assessment made by the Examiner.

The Examiner, however, states that "[Jukema] ... discloses the evidence for a synergistic effect of calcium channel blockers ... with lipid-lowering therapy in retarding progression [of] coronary atherosclerosis ..." The examiner continues, "[t]he lipid-lowering agents employed therein in the combined therapy are HMG-CoA reductase inhibitors such as pravastatin ..." Continuing, "[o]ne of calcium channel blockers employed therein is amlodipine ..."

The Examiner next addresses the Merck Index stating that the Index "lists atorvastatin as a HMG-CoA Reductase Inhibitor, belonging to the same therapeutic group as Pravastatin ..."

The Examiner concludes by stating, "[i]t would have been obvious to one of ordinary skill in the art at the time the invention was made to employ amlodipine besylate with atorvastatin metabolites in a [pharmaceutical] composition."

In order to establish a *prima facie* case of obviousness, "there must be some suggestion or motivation, either in the references themselves or in the knowledge generally available to one of ordinary skill in the art, to modify the reference or to combine reference teachings. Second, there must be a reasonable expectation of success. Finally, the prior art reference (or references) must teach or suggest all of the claim limitations." M.P.E.P. §2143, see also, *In re* Vaeck, 947 F.2d 488, 20 USPQ2d 1438 (Fed. Cir. 1991).

Moreover, it is axiomatic in patent law that if an independent claim defines allowable subject matter then the claims depending therefrom also define allowable subject matter. *In re* Fine, 837 F.2d 1071, 5 USPQ2d 1596 (Fed. Cir. 1988), and Hartness International, Inc. v. Simplimatic

Engineering Co., 819 F.2d 1100, 1108, 2 USPQ2d 1826, 1831 (Fed. Cir. 1987). Given that the rejected claims depend from base claims and those independent claims define allowable subject matter, then the claims at issue must necessarily define allowable subject matter. The reasons for allowability of the base claims are set forth herein.

Neither Davison nor Bjorge, alone or in combination with either Jukema and/or the Merk Index, provides a sufficient case of *prima facie* obviousness. The cited references neither teach nor suggest the presently claimed invention. For example, these cited references alone or in combination do not teach or suggest a combination therapy using amlodipine and an atorvastatin metabolite for synergistically inhibiting lipid peroxidation in LDL or membrane lipid bilayers, and/or for use as an antioxidant.

The Examiner states that neither Davison nor Bjorge alone teach or suggest the present invention. However, the Examiner employs Jukema *et al.* as a reference that allegedly supplies that which is deficient in Davison and Bjorge. The Examiner contends that Jukema *et al.* disclose evidence for a synergistic effect of calcium channel blockers with lipid-lowering therapy.

In fact, Jukema does not provide evidence for a synergistic effect. In Dr. Jukema's declaration, referring to his own paper which is cited by the Examiner, he states that "we did not conduct any original experiments for this analysis of the REGRESS study, such as data showing a *synergistic effect* using a combination therapy comprising amlodipine and atorvastatin (or any of their analogs)." [*italics added*] See paragraph 4 of Jukema Declaration. Therefore, an inference cannot be drawn from Jukema's work that a combination of amlodipine and atorvastatin has any synergistic properties. Hence, such an inference is improper to be imported into an argument in support of a case of *prima facie* obviousness.

The presently claimed invention has been amended to clarify that the invention is directed toward a combination of amlodipine and hydroxylated atorvastatin, wherein said combination has synergistic properties. See, *e.g.*, FIG. 1, additionally, refer to Dr. Mason's previously submitted Declaration. Based on Dr. Jukema's Declaration, the cited reference used to construct a *prima facie* case of obviousness against the presently claimed invention, *i.e.*, his own paper, does not provide for any evidence of synergy between amlodipine and hydroxylated atorvastatin. Again, to impute any synergistic property based on Jukema would be improper and untenable.

Further, Dr. Jukema states in paragraph 7 of his declaration, "that our paper did *not* mention the use of atorvastatin as a combination agent or specifically suggest the use of amlodipine in combination with the HMG-CoA reductase inhibitor atorvastatin." [*italics added*] In fact, in paragraph 6 Jukema declares "that the REGRESS trial was *not* designed to evaluate combination therapy, specifically, a therapy combining a CCB with an HHMG-CoA reductase inhibitor." [*italics added*] These declarations clearly indicate that Jukema's paper cannot be used to supply the deficiencies present in Davison and Bjorge as correctly pointed out by the Examiner.

It must be appreciated that Jukema *et al.* is a retrospective analysis of a clinical investigation and it can be argued that it teaches away from our invention as it shows that patients on the combination of a CCB and statin actually had more adverse clinical events. Jukema *et al.* states that patients receiving the combination of a calcium channel blocker with the HMG-CoA reductase inhibitor pravastatin did *not* experience fewer adverse clinical events as compared to patients that were not on calcium channel blocker therapy with pravastatin. In fact, it was reported in the article that patients that were being simultaneously treated with both a calcium channel blocker and pravastatin actually had more cardiovascular complications (*e.g.*, more revascularization procedures) than patients not taking the treatment combination. Although the increase in adverse events with the drug combination was not statistically significant, it further argues against a synergistic therapeutic benefit based on this retrospective nonexperimental analysis. Jukema *et al.* recognized the combination of a calcium channel blocker with pravastatin improved anatomical changes in certain coronary vessels, as measured with angiographic approaches, but this did not correlate with an improvement in clinical outcomes - this is in contrast to Examiner's argument.

Dr. Jukema addresses this very point in his Declaration as well. In paragraph 14 he declares, "that the improved anatomical changes *i.e.*, angiographic changes in lumen size in certain coronary vessels following combination therapy did *not* correlate with a significant reduction in clinical events." [*italics added*] Further in paragraph 15 he declares, "that the reason for the anatomical changes may be due to confounding factors and may not directly be related to the combination treatment." In paragraph 16 he declares, "that the absence of a correlation between significant changes in lumen size of the coronary artery (measured by angiography) clinical events may be due to the low number events in REGRESS but is also consistent with other angiographic-based trials at the time ..." Clearly, as evidenced by the authors own words, there is no correlation between improved anatomical features and combination therapy as claimed in the present invention.

The Examiner also cites the Merck Index stating that the Index "lists atorvastatin as a HMG-CoA Reductase Inhibitor, belonging to the same therapeutic group as Pravastatin ..." (Applicant assumes that the Examiner makes this statement due to the inclusion of pravastatin in the Jukema paper.) Dr. Jukema in his Declaration states "that I did not present data nor discuss that a synergistic effect observed with amlodipine and atorvastatin metabolite could be reproduced using other statins." Dr. Jukema by this declaration is informing us that one should not assume that it can be assumed that pravastatin will have the same effect as the atorvastatin metabolite when combined with amlodipine. Moreover, FIG. 5 illustrates this point where other statins where used kin combination with amlodipine and a synergistic effect was not observed. Clearly, this suggests that there is a unique relationship between atorvastatin and amlodipine and that one cannot generalize at this time as to a combination of classes producing synergistic effects.

Clearly, as supported by the above discussion, there is nothing in Jukema that teaches or suggests the presently claimed invention. Moreover, Jukema lacks any motivation to one skilled in the art to combine either Davison, Bjorge and/or the Index with Jukema. Significantly, there is no reasonable expectation that if one were to combine these references that one would arrive at the presently claimed invention.

In conclusion, in view of the above amendments and remarks, Applicant respectfully requests the Examiner find the pending claims in condition of allowance and, therefore, issue a Notice of Allowance. Although no additional fees are required, please charge any underpayment of fees to or credit any overpayment of fees to Deposit Account No. 03-2410. The Examiner is invited to call the undersigned attorney at (617) 854-4237 should he determine that a telephonic interview would expedite prosecution of this case.

Respectfully submitted,

Stephen J. Gaudet, Ph.D.
Attorney for Applicant
Reg. No. 48,921

Date: July 15, 2004

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of: Mason, R.P. Group Art Unit: 1617

Application Serial No.: 10/033,149 Examiner: Jiang, S.A.

Filed: October 19, 2001

For: Synergistic Effects of Amlodipine and Atorvastatin Metabolite as a Basis for Combination Therapy

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Assistant Commissioner of Patents
Washington, D.C. 20231

Sir:

DECLARATION UNDER 37 CFR 1.132

I, R. Preston Mason, Ph.D., declare that I have the following positions, education and qualifications in the field of biomedical science:

1. I am the President and Founder of Elucida Research in Beverly, Massachusetts, an independent research laboratory dedicated to innovative biomedical research since 2000.

2. I am a faculty member in the Department of Medicine at Harvard Medical School in Boston, Massachusetts. I am also a senior research scientist at the Harvard – affiliated facility, Brigham and Women's Hospital in Boston.

3. I received my bachelor's degree in biology (*Summa Cum Laude*) from Gordon College in Wenham, MA in 1985. I received the Sarah Ball Award for the highest G.P.A. among all students majoring in science in 1984. Following graduation, I received a scholarship to attend the University of Connecticut School of Medicine (MD/PhD predoctoral fellowship) where I received a Ph.D. in 1985 in Biomedical Sciences with a concentration in Cell Biology and Biophysics. After graduation, I

received two consecutive post-doctoral fellowships from the American Heart Association, Connecticut Affiliate.

4. I joined the faculty at the University of Connecticut School of Medicine in 1991 as an assistant professor of radiology and an independent investigator in the Biomolecular Structure Analysis Center. In 1994, I moved to Pennsylvania as I was invited to join the faculty of MCP Hahnemann University School of Medicine (now part of Drexel University) as an associate professor of psychiatry, biochemistry and medicine until 2001 when I returned to New England.

5. My medical research interests broadly include membrane biophysics, oxidative stress, vascular biology and molecular pharmacology. A frequent lecturer at national and international meetings and academic seminars, I have written or coauthored more than 80 full-length articles and book chapters related to my research in journals such as *Circulation, Biochemistry, Journal of Biological Chemistry, Journal of Lipid Research* and *Journal of the American Medical Association*. This past year, I was invited to present my research in cardiovascular pharmacology at the annual meetings of the American Heart Association and the American College of Cardiology. I also co-chaired a scientific session at the American Heart Association annual meeting in 2003.

6. I have served as an expert scientific reviewer for the National Institutes of Health and the American Heart Association, Southeastern Pennsylvania Affiliate, for several years.

I further declare:

(1) that I am currently working as an active researcher in the area of vascular physiology/pharmacology and study calcium channel blockers such as amlodipine and HMG-CoA reductase inhibitors, including atorvastatin and its active metabolite;

(2) that I have supervised experiments, as explained below, in my laboratory in which I observed a strong *synergistic* antioxidant effect with amlodipine when combined with the active, o-hydroxy metabolite of atorvastatin;

(3) that these experiments utilized the materials and methods herein described. For clarity, the following abbreviations are used: BHT: butylated hydroxy toluene; ddi H_2O: double deionized water; LDL: low density lipoproteins; PBS: phosphate buffered saline; TBA: thiobarbituric acid; TBARS: thiobarbituric acid reactive substances; TCA: trichloroacetic acid; and EDTA: ethylenediaminetetraacetic acid;

(4) that the materials used in these experiments specifically were: Human LDL from Calbiochem; Phosphate buffered saline (pH = 7.4) from Sigma; Sephadex G-25M (PD-10) columns from Pharmacia Biotech; $CuSO_4$ (prepared in ddi H_2O) from VWR Scientific; Trichloroacetic acid (5%) from VWR; Butylated hydroxytoluene (20 µM in redistilled ethanol) from Sigma; Thiobarbituric acid (0.5% in ddiH_2O) from Sigma; Protein standard (bovine serum albumin at 1 mg/ml) from Sigma; BioRad Protein Assay Dye Concentrate from BioRad Laboratories; Amlodipine besylate stock in redistilled ethanol. (F.W. = 567) from Pfizer Inc.; and Atorvastatin o-hydroxy metabolite stock in redistilled ethanol (F.W. = 638) from Pfizer Inc;

(5) that the methods included the following preparation of human LDL. LDL came in a solution containing EDTA, a common preservative. The EDTA was removed by gel filtration with PD-10 Sephadex G-25M columns. The LDL was made up to a volume of 2.5 ml using PBS (N_2 purged). The PD-10 column was

equilibrated with 25 ml of PBS column. The LDL was loaded on the column and then washed with PBS and the eluant was collected as 3 fractions. These 3 fractions were then analyzed for protein using the BioRad Assay;

(6) that the protein content of the human LDL was measured using the Biorad Protein Assay. The protein standards obtained from Sigma were diluted to obtain four standards at fixed concentrations of 0, 1, 5 and 10 µg/ml. Three samples were also run, consisting of the various LDL fractions (20 µL of each fraction was tested). To each standard and sample, 300 µL of BioRad dye concentrate was added. The content of the tubes were mixed before allowing the sample to incubate for 5 minutes at room temperature. The absorbance of the solution was read at 595 nm;

(7) that the equation of the standard protein curve was: $y = 0.0422n + 0.5183R^2 = 0.9956$. Based on this calculation, the protein concentrations of the 3 LDL fractions were: fraction 1: 16.47 µg/ml; fraction 2: 368.01 µg/ml; and fraction 3: 65.05 µg/ml. Fraction 2 was then used for the lipid peroxidation assays. Fraction 2 was then diluted with PBS to obtain a final protein concentration of 50 µg/ml and this was the stock LDL used in all of our assays;

(8) that the following samples were prepared: LDL (50 µg/ml) treated with vehicle alone; LDL + Atorvastatin Metabolite (100 nM); LDL + Amlodipine (2.5 µM); LDL + Atorvastatin Metabolite (100 nM) + Amlodipine (2.5 µM); and LDL + Lovastatin (100 nM) + Amlodipine (2.5 µM). To prepare the samples, LDL (50 µg/ml) was incubated with 10 µL of ethanol (vehicle) in the control samples or with the respective drugs prepared in a 10 µL volume of ethanol at 37°C for 30 minutes. LDL oxidation was then initiated by the addition of 10 µL of $CuSO_4$ (10 µM final concentration). At the time the $CuSO_4$ is added is time = 0 hours. At specific time points (1.5 hr, 2.0 hr), a 100 µL aliquot was removed from each tube and tested for the extent of lipid peroxidation;

(9) that the extent of LDL oxidation in the absence or presence of the drugs was measured by TBARS analysis. The LDL-containing sample (100 µL) was

added to a test tube containing 10 µL of 5% TCA, 10 µL of BHT (20 µM) and 1 ml of TBA reagent. The tube was capped and then heated at 80°C for 30 minutes on a heating block. The sample was cooled and the absorbance read at 532 nm. The molar-extinction coefficient for TBARS is $1.56 \times 10^5 M^{-1} cm^{-1}$; and

(10) that the significance of differences between results from the various experimental conditions was tested using the two-tailed Student *t*-test, multiple analyses (n = 9-10) were done for each experimental condition, and significance was accepted at a level of 0.05.

I further declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

RPMum — July 15, 2004

(Name) **(Date)**